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Investigation of autologous chondrocyte transplants and thereof isolated cells

H. Zwickl1, P. Holzmann1, T. Luksh1, E. Niculescu-Morza2, S. Nehrer3; 1Centre For Regenerative Medicine, Department For Clinical Medicine And Biotechnology, Danube University Krems, Krems, Austria, 2Department For Orthopedics, Landesklinikum Krems, Krems, Austria, 3Abteilung Für Orthopädie, Landesklinikum Krems, Krems, Austria, <Center For Regenerative Medicine, Danube University Krems, Krems, Austria>

Purpose: ArthrokInetics produces (autologous) transplants (CaReS®) for repair of large-area cartilage defects (2-10 cm²) by isolating chondrocytes from a patient’s biopsy and combination with a collagen type I matrix. 11-15 days after biopsy it is ready for implantation. Before matrix embedment, chondrocytes are tested concerning vitality and chondrocyte-specific synthesis performance (e.g. collagen II). We investigated transplant-embedded chondrocytes at the time of implantation as well as proliferation and sGAG synthesis after isolation from the transplant and during subsequent cultivation.

Methods and Materials: Transplants of 5 different patients were investigated. Chondrocytes were isolated by enzymatic digestion of biopsy tissue, concentrated and seeded in 6-well plates (2x10⁶ cells/well). Cells were cultivated in DMEM/F12 (supplemented with ascorbic acid and streptomycin/penicillin) either without or with 10% FCS. sGAG were quantified according to the DMMB method (Barbosa et al.).

Results: The cell number of the different ready-to-use transplants varied between 2,13X10⁵ and 1,3X10⁶ cells/g transplant, the amount of transplant-associated sGAG between 7 and 29 µg/g transplant. The amount of sGAG/10⁵ cells ranged from 2 to 12 µg, whereupon proliferation-promoting conditions led to an increase of the originally seeded cell number of 66% to up to 486%.

Conclusions: Chondrocytes in transplants synthesized sGAG during the incubation period regardless of the matrix-embedded implant. They were vital after isolation from the transplant and proliferated and retained their sGAG synthesis performance under proliferation-promoting culture conditions during the observed time period.

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High resolution multiparametric MR compression study of human articular cartilage at 3 Tesla using unique compression device

V. Juras1, M. Bittsansky1, M. Jajdusova2, S. Trattnig1; 1Department Of Radiology, MR Centre - High Field MR, Vienna, Austria, 2Department Of Orthopaedic Surgery, University of Tromsø, Tromsø, Northern Norway, 3Department Of Clinical Chemistry, University hospital of Northern Norway, Tromsø, Norway

Purpose: The purpose of this study was to use special designed compression device adapted to a micro-gradient system to study the effects of mechanical compression of human cartilage explants on T1, T2 and apparent diffusion coefficient (ADC) by application of a highly accurate and localized compression of articular cartilage.

Methods and Materials: Cartilage samples were prepared from joints of 10 patients, who underwent a total knee joint replacement, with 10x10x3 mm in dimension. Study was performed on a Bruker 3T Medspec whole-body scanner using BGA-12 micro-gradients with a special designed compression device built for this gradient system. This equipment allows to move the compressive piston with accuracy of 10.735, 220.879, 453.036, 724.697, 957.897. Equipment for cartilage compression evaluation is used approach is that data obtained from a micro-imaging system that addition of growth factors may help to the proliferative potential of some catabolic molecules. Cells apoptosis decreased during the culture period evaluated.

Results of this study showed that T1, T2 and ADC are related to the pressure loaded onto a cartilage tissue. In case of cartilage thickness change of 15%, ADC decreased for 10.15%, T1 increased for 22.6% and T2 decreased for 13.97%.

Conclusions: Equipment for cartilage compression evaluation is feasible for studying influence of pressure on cartilage tissue with high accuracy of localization and loading control. The advantage of used approach is that data obtained from a micro-imaging system provide high-resolution images and precise localization of every compared pixel.

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Rationale for the use of tissue engineering in early cartilage lesions in osteoarthritis

B. Grigolo1, G. Lisigoni1, L. Roseti1, C. Cavailllo2, G. Desando3, A. Facchini2; 1Laboratorio Di Immunologia E Genetica, Istituti Ortopedici Rizzoli, Bologna, Italy, 2Banca Del Tessuto Muscoloscheletrico, Istituti Ortopedici Rizzoli, Bologna, Italy, 3Laboratorio Di Immunologia E Genetica, Istituti Ortopedici Rizzoli; Università degli Studi di Bologna, Bologna, Italy

Purpose: To investigate whether autologous chondrocyte transplantation could be used to treat early cartilage lesions in patients with osteoarthritis exploiting the regulatory effect of a hyaluronan-based scaffold.

Methods and Materials: Human chondrocytes were isolated from articular cartilage obtained from patients with early-onset knee osteoarthritis. The cells were expanded in monolayers and seeded on a hyaluronan-acid derivative scaffold already used in cartilage repair. Constructs and surnantants were collected and analyzed at 1, 3, 7, 14 and 21 days after seeding. Immunohistochemical analysis for CD44 and caspase was carried out on paraffin embedded sections.

Conclusions: The hyaluronan-based biomaterial used in this study acts on chondrocyte metabolism downregulating catabolic pathways. The ability to reduce the expression of molecules involved in cartilage degenerative pathways by chondrocytes indicates its use also in the transplantation therapeutic strategy to treat early lesions in patients with osteoarthritis.

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Morpho-functional characterization of human articular chondrocytes expanded under different cell culture conditions

J. Elvenes1, I. Martinez2, S. Bendiksen3, O. Johansen4; 1Department Of Orthopaedic Surgery, University of Tromsø, Tromsø, Norway, 2Department Of Clinical Chemistry, University hospital of Northern Norway, Tromsø, Norway

Purpose: Currently there is no consensus on the optimal culture method for autologous articular chondrocytes intended for cartilage repair surgery. In this study, we evaluated the changes in the morphology and function of the chondrocytes when altering the culture conditions during cell expansion.

Methods and Materials: Human articular chondrocytes where exposed to different culture conditions. The control group was treated with the standard culture method including 10% human serum and atmospheric oxygen levels. The varying factors where the presence of CD 44 receptor increased over time showing the highest positivity at day 21. Immunohistochemistry for caspase-3 displayed many positive cells at 1 day but on day 21 the cells were almost negative. Decreased levels of chemokine and nitric oxide were observed in the surnantants of chondrocytes grown onto the scaffold. Real-Time PCR showed that the cells expressed the specific differentiated phenotype and downregulated the expression of some catabolic molecules. Cells apoptosis decreased during the culture period evaluated.

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